

CALIFORNIA ENVIRONMENTAL PROTECTION AGENCY
DEPARTMENT OF PESTICIDE REGULATION
MEDICAL TOXICOLOGY BRANCH

SUMMARY OF TOXICOLOGY DATA

FENARIMOL

Chemical Code # 001980, Tolerance # 421
SB 950 # 285

10/1/86

Revised 5/28/87, 9/13/88, 3/19/90, 9/9/93

I. DATA GAP STATUS

Chronic rat:	No data gap, no adverse effect
Chronic dog:	No data gap, no adverse effect
Oncogenicity rat:	No data gap, possible adverse effect
Oncogenicity mouse:	No data gap, no adverse effect
Reproduction rat:	No data gap, no adverse effect
Teratogenicity rat:	No data gap, no adverse effect
Teratogenicity rabbit:	No data gap, no adverse effect
Gene mutation:	No data gap, no adverse effect

Chromosome effects: No data gap, no adverse effect

DNA damage: No data gap, no adverse effect

Neurotoxicity: Not required at this time.

NOTE: Toxicology one-liners are attached

** indicates acceptable study

Bold face indicates possible adverse effect

Toxicology Summary revised by M. Silva, 3/19/90 & 9/9/93.

FILE NAME: T930909

Reconciled through volume #: 070 and record #: 991740 & 089208.

NOTE: ONE-LINERS PROVIDE A GENERAL SUMMARY. SEE WORKSHEETS FOR COMPLETE REVIEWS.

TOXICOLOGY ONE-LINERS AND CONCLUSIONS:

CHRONIC OR ONCOGENICITY STUDIES, RAT

**** 017-020 037237-037240** (The principal long-term rat study) "Twenty-four month chronic oral toxicity of EL-222 (56722) in rats", [Studies R-405 and R-415]. Lilly Research Labs, 4/78. [Other relevant studies include 035:42863; 021/022:037244-037245; 020/021:037241-037242; 034:042859; and 036:050760]. Dosages in principal study = 0, 50, 130, and 350 ppm in diet. Wistar-derived rats: total of 120/sex in controls and 80/sex/group in treated groups. Fenarimol, tech., 97.9%. **Possible adverse effect:** Apparent NOEL = 130 ppm [minor increase in hepatocellular adenomas (not significant by most 2-way statistical comparisons), and moderate to severe fatty change in liver at 350 ppm]. Incidence of hepatocellular adenomas from studies R-405 and R-415 in controls through increasing doses was: Males- 0/116, 0/78, 1/77, and 3/79; Females- 0/116, 0/80, 0/80, and 2/79. **This study, supported by other relevant studies listed, is now acceptable, and fills the combined rat (chronic-oncogenicity) data requirement.** C. Aldous, 4/21/86, 5/12/87; conclusions re-examined by C. Aldous in conjunction with reviews of additional data in Vol. 044 (see below), 8/24/88 and 8/25/88.

003 991736 (Identical to first 224 pages of R-405/R415, DPR Volume/record#: 017-020/037237-037240).

NOTE: EPA initially determined that liver tumors of the rat study (017:037237) were a treatment effect. The Federal Register of March 5, 1986 (Vol. 51, No. 43, pp. 7567-7568) read: "The compound demonstrated an oncogenic effect of significant increase of hepatic adenomas and hyperplastic nodules at 17.5 mg/kg bw/day". Later, EPA changed its conclusion and determined that fenarimol was not indicative of oncogenicity (see Vol. 046, no record number). The statement was entitled: "Pesticide tolerances for fenarimol" from 10/30/86 Federal Register 51:39660-39662. The original EPA evaluation had lumped hyperplastic nodules with tumors (adenomas and carcinomas), and had grouped male and female responses together.

When sexes were considered separately, and when the hyperplastic nodules were removed from the tumor incidence count, EPA found no statistically significant differences for either sex. (C. Aldous, 8/29/88, no separate written review).

044 063781 "Assessment of the potential oncogenicity of fenarimol". [Several rat and mouse long-term studies were discussed, however the study of greatest importance was rat combined study 017:037237]. Report was prepared for Elanco Products Co. Major studies being discussed were conducted at Toxicology Division of Lilly Research Laboratories, Greenfield, Indiana. Date of report (044:063781) was 7/1/86. This report reviewed the bases for dose justification for the above rat study, and for the mouse oncogenicity study (014:037233). The report also reviewed statistical data, which were presented as indications that there was no treatment effect on rat hepatocellular tumor incidence. Mutagenicity data, which were generally negative, were cited as additional evidence that there was no oncogenicity treatment effect. No change in status of studies is indicated, however statistical analyses and other data may be useful for subsequent risk assessment. C. Aldous, 8/24/88.

044 063787 "The incidence of hepatic nodules and neoplasms in two-year old control Wistar rats and Wistar rats from long-term studies with Fenarimol (EL-222)". Toxicology Division, Lilly Research Laboratories historical incidence data for hepatocellular lesions: hyperplastic nodules, adenomas, and carcinomas. Supplementary report dated March, 1986. Incidence of hepatocellular hyperplastic nodules, adenomas, and carcinomas in males (out of 1380) was 3, 6, and 3. Respective incidence in females (out of 1401) was 8, 2, and 3. Cases of > 1/sex/study were limited to 2/60 males in one study with adenomas, 2/90 males in one study with nodules, 4/60 and 2/55 females with nodules in two studies. These data are of potential value for risk assessment. C. Aldous, 8/25/88.

021 037243 Brief synopsis of historical incidence data more completely described in 044:063787, above.

035 042863 "Eighteen-month chronic oral toxicity of EL-222 (56722) [Fenarimol] in [Wistar] rats" [Study 435]. Lilly Research Labs, May 1978. Fenarimol, tech. 0, 50, 130, and 350 ppm

in diet. Apparent NOEL = 50 ppm (based on body weight decrements in 130 and 350 ppm males). Liver enlargement in both sexes at 350 ppm, fatty liver in 350 ppm females. Not complete, not acceptable, but useful information. Aldous, 8/8/86.

021-022 037244-037245 "A chronic toxicity/oncogenicity study in Wistar rats maintained on diets containing low concentrations of Fenarimol for two years" (Study R07781). Lilly Research Laboratories, Toxicology Div., May 1985. Fenarimol, technical, 98.9% purity. 0, 12.5, 25, and 50 ppm in diet. NOEL = 50 ppm (HDT). Not acceptable, not complete (but no more data required of this study). Useful data. C. Aldous, 2/20/86.

020-021 037241-037242 "A low-dose chronic toxicity/oncogenicity study in Wistar rats maintained on diets containing Fenarimol for two years" (Study R06479). Lilly Research Labs, Nov. 1982. Fenarimol, tech., 96.7% purity. 0, 12.5, 25, and 50 ppm, dietary. NOEL = 25 ppm (increased incidence of slight fatty change in liver). Not complete (No more data needed--a more recent study, 021/022:037244-037245, is already done). Not acceptable (unacceptable losses to disease). C. Aldous, 2/19/86.

036 050760 "A one-year toxicity study with EL-222 in the rat" (Study R-715). Lilly Research Labs, June, 1978. Fenarimol, tech. (97.9%). 0, 50, 130, and 350 ppm in diet. Wistar-derived rats were F0 parents from a reproduction study. Apparent NOEL = 130 ppm (Slight increase in relative liver and kidney weights of females). Study limited in value: too short duration for oncogenicity evaluation. Unacceptable to fill chronic effects data gap, but useful information. Study cited as ancillary information to 017-020:037237-037240 in C. Aldous review, 5/12/87.

034 042859 "Initiation and promotion study of Fenarimol on rat liver carcinogenesis". Naylor Dana Institute for Disease Prevention, Nov. 22, 1985. Fenarimol (Lot B30-C69-220, approx. 97.9% purity). Fenarimol administered to young male F344 rats at 350 ppm in diet for 8, 12, or 20 weeks: possible interaction with tumor initiator (FAA) and with promoter (phenobarbital) was investigated. Results: No evidence of initiation nor promotion due to fenarimol. **Acceptable** ancillary study to principle rat combined study (017/020:037237-037240), on basis

of additional information in rebuttal document (Vol. 036, pp. 51-53). C. Aldous, 8/8/86,
5/26/87.

003 991736 (Identical to first 225 pp. of R-405/R415 (017-020:037237-037240))

CHRONIC, DOG

** 032 042702 "A one year chronic oral toxicity study of Fenarimol in dogs with a three month recovery period". Study No. D02683. Lilly Research Labs, May, 1985. Fenarimol, tech., 96.7%. 0, 1.25, 12.5, and 125 mg/kg/day by capsule. No adverse effects indicated. NOEL = 12.5 mg/kg/day (increased liver weight, increased p-nitroanisole O-demethylase activity and mild bile stasis in liver, and increased alkaline phosphatase). Additional information in 036:050759 also pp. 22-27, (no record No.). **Study acceptable.** C. Aldous, 8/19/86, 5/18/87.

036 050759, (pp. 62-69). Part of response to EPA and DPR concerns about enlarged mammae in study 032:042702. Review by Dr. Patrick W. Concannon, Dept. of Physiology, Cornell Univ. "Review of reproductive activity and mammary gland activity in female dogs in Lilly Research Study D02683 of Compound 56722". Observations of enlarged mammae do not appear to represent a treatment effect.

034-036 042858 Response to EPA review of 032:042702. This information considered in 8/19/86 review.

ONCOGENICITY, MOUSE

** 014-016 037233-037236 "Twenty-four month chronic oral toxicity of EL-222 (56722) in mice" [Studies M-9135 and M9145]. Oncogenicity, mouse, [832]. Lilly Research Labs, 7/31/78. Fenarimol, tech., 97.9% purity. 0, 50, 170, and 600 ppm in diet. NOEL = 170 ppm: Slight fatty metamorphosis in male livers. Acceptable (fundamental issue preventing earlier acceptance was lack of defensible MDT: acceptance was based on consideration of criteria in 1986 EPA position document on MTDS in oncogenicity studies). C. Aldous, 2/14/86, 5/14/87.

002 991728 Exact duplicate of first 160 pages of 014-016:037233-037236, above (through table summarizing necropsy data). C. Aldous, 7/24/85.

002 991735 A 1981 rendition of final report 014-016:037233-037236, above (through table summarizing necropsy data--excludes individual data). Data from the paired studies are combined in tables, and format of this report is more convenient for evaluation than the 1978 final report. C. Aldous, 7/24/85.

REPRODUCTION, RAT

Summary of rat reproduction data, considering 024 037258 and the many associated studies of fenarimol effects on reproduction: The major reproductive findings in rats (male infertility, dystocia and related effects in females, hydronephrosis in fetuses and pups) do not indicate "possible adverse health effects" of consequence to man. Male infertility and dystocia in dams seem to be relatively species specific. Important physiological differences between man and rat include: (1) The two species depend on different testosterone-derived effector molecules to moderate early stages of male behavioral sexual development. In **rats, mice, and hamsters**, aromatase forms the effector molecule (estradiol), which is essential to central nervous system organization of male behavioral patterns. Aromatase activity is inhibited by fenarimol; and this inhibition appears to account for some or all reproductive toxicity observed in rats. Rabbits and primates are less dependent or independent on aromatase activity in the mediation of testosterone on central nervous system sexual behavioral development (see 003 991732, p. 563), thus the effects observed in rats (at least males) are unlikely to be relevant to man. (2) The hormonal changes observed in female rats, which are associated with dystocia, are at least in part irrelevant to humans (i.e. fenarimol inhibits the normal drop in progesterone levels, which is an important step in initiating parturition in rats. On the other hand, a drop in progesterone levels is **not** essential to initiate parturition in humans). Negative findings for reproductive effects at appreciably high dose levels in guinea pig and rabbit reproduction studies are added indications that reproductive effects seen in rats represent species differences. (3) The slight to moderate degree of hydronephrosis apparently elicited by fenarimol does not appear to be physiologically important (no damage to parenchymal tissues), and hydronephrosis is not definitively observed except at comparatively high dosages. C. Aldous, 9/9/88. **NOTE:** A

pesticide tolerance for fenarimol was the subject of a Federal Register entry on March 5, 1986 (Vol. 51, No. 43, pp. 7567-7568). EPA reviewers accepted the registrant's assertion that reproductive effects noted in this general review for Wistar rats are species-specific. The reproductive NOEL was established as 35 mg/kg/day based on a guinea pig study (presumably study 025:037297, discussed below).

** 024 037258 "A multi-generation reproduction study with EL-222 in the rat". (Lilly study designations: R-715, R-1345, R-956, and R-966) Lilly Research Labs, Nov., 1977. Fenarimol (EL-222), Lot No. B30-C69-220 (97.9%). 0, 50, 130, and 350 ppm in diet. Determinations of NOELs and hazard evaluation indicated here were based on a large body of studies which characterize reproductive effects of fenarimol. Overall reproductive effects NOEL = 25 ppm (reduced fertility, due to decreased copulatory behavior by males; occasional dystocia in dams down to 50 ppm, which was attributed by investigators to test article) based on consideration of follow-up study 024:037259. There was weak evidence in 024:037259 of increased unilateral hydronephrosis with an apparent NOEL of 12.5 ppm; however the primary study (024:037258) suggests a substantially higher NOEL for hydronephrosis. The profile of reproductive effects at 350 ppm was: reduced liveborn litter sizes, increased gestation time or delayed onset of parturition, apparent dystocia-related increase in fetal deaths, slight decrease in pup survival, increased incidence of hydronephrosis in weanlings. Study was initially classified not acceptable because of significant variances from current guidelines and because reproductive effects were not adequately explored. This study, with information previously submitted, is **accepted** on receipt of two additional supplementary reports requested by DPR (044 037258 and 037259). DPR **removes "possible adverse effect" flag** on evaluation of the collective reproductive effects data now available (see summary paragraph above). Aldous, 3/21/86, 5/20/87, 9/9/88.

044 063788 (Supplementary study relates to principal reproduction study 024 037258). Title of supplementary study: "Inhibition of plasma estradiol levels by fenarimol (EL-222) in rats prior to parturition". Lilly Research Laboratories (Indianapolis), 2/85. 0 or 350 ppm fenarimol administered in feed, apparently from time of breeding through pregnancy. Plasma estradiol was monitored on days 19-22. Treated animals had consistently reduced estradiol,

statistically significantly reduced from day 19 AM through Day 20 AM. Explanation: Normally a sharp rise in estradiol occurs a few days before parturition and continues through the time of parturition. The demonstrated delayed rise in estradiol levels is indicative of delayed luteolysis and is consistent with delayed parturition and associated dystocia in rats observed with fenarimol at this dosage level. These results are consistent with (but do not prove) a reduction in aromatase activity within the corpora lutea. Report is sufficiently complete to serve as a meaningful supplementary study. C. Aldous, 8/26/88.

044 063789 (Supplementary study relates to principal reproduction study 024 037258). Title of supplementary study: "Summary of gross and histopatho-logical findings in the reproductive tracts of individual male and female rats". Lilly Research Laboratories (Greenfield, IN), 9/87. Individual gross and histopathological findings were presented for F1 adults of study 024:037258. No findings appeared to correlate with male or female reproductive effects observed at that dosage. (There was an apparent slight increase of observations of abscesses or purulent material in the uteri of the high dose group, however this is unlikely to represent a treatment effect). C. Aldous, 8/26/88.

024 037259 "A second multi-generation reproduction study with EL-222 (Lilly compound 56722) in the rat". Lilly Research Labs, July 1978. Fenarimol, tech. (97.9%). 0, 12.5, 25, and 50 ppm in diet. NOEL for reproductive effects in pups = 12.5 ppm (hydronephrosis at 25 ppm and above). NOEL for parental animals = 25 ppm (occasional dystocia at 50 ppm). Significant reduction in fertility at 50 ppm. This study is ancillary to 024:037258, above: Study was not designed as an independent reproduction study. Parental animals generally not necropsied. (Aldous, 3/26/86).

024 037262 "A cross-over fertility study with EL-222 in the Wistar rat" (Study R08379). Lilly Research Labs, June 1980. Fenarimol, 97.9%. 0 and 35 mg/kg/day by gavage. No NOEL established. Effects at 35 mg/kg/day (gavage) were: dystocia in dams with prolongation of gestation, failure of males to copulate, increased stillbirths and decreased litter sizes when dams are treated. Early neonatal survival of such dams significantly reduced. An ancillary reproduction study only. (Aldous, 3/28/86)

024 037263 "A cross-over reproduction study with EL-222 (Lilly Compound 56722) in the rat" [Study R-286]. Lilly Research Labs, April, 1980. Fenarimol, 97.9%. 0 and 35 mg/kg/day by gavage. No NOEL established. Treated males had reduced copulation. Treated females had smaller litters and increased stillbirths. Ancillary study, designed as a supplement to other reproduction studies. (Aldous, 3/28/86).

024 037264-037265 "Additional cross-over reproduction studies with EL-222 (Lilly Compound 56722) in the Wistar and the Sprague-Dawley rat" [Wistar Rat, Study R-57; Sprague-Dawley Rat, Study R-67], Lilly Research Labs, April 1980. Fenarimol, 97.9%. Doses of 0 or 35 mg/kg/day by gavage in 1-generation cross-over design. No NOELS established. At 35 mg/kg/day the following were seen: Failure to copulate in males, dystocia and increased incidence of stillborn pups with smaller liveborn litter sizes. Females had increased luteinizing hormone and decreased prolactin levels when pups were weaned. An ancillary reproduction study. (Aldous, 3/31/86).

036 (no Record number, but pp. 28-33 near beginning of volume); Main text of 11/25/86 registrant rebuttal, identifying many studies which collectively characterize reproductive effects of fenarimol (addressed in C. Aldous Worksheet of 5/20/87).

036 050760 "A one-year toxicity study with EL-222 in the rat" (Study R-715). Attachment 3 of 11/25/86 registrant rebuttal, a "chronic" study and extension of the "Core" (024:037258) reproduction study). No adverse reproductive tissue or behavioral effects noted (addressed in C. Aldous Worksheet of 5/20/87).

036 050762 "Fenarimol--Relevance of reproduction findings to man"). Attachment 4 of 11/25/86 registrant rebuttal, a summary of findings in numerous reproductive and teratology studies, indicating that the reproductive hazards of fenarimol have been adequately addressed, and summarizing many studies to support the conclusion (addressed in C. Aldous Worksheet of 5/20/87).

003 034781-034783, 991732 "An overview of the effects of EL-222 (Fenarimol) on reproduction in laboratory animals and the relevance of these effects to risk assessment in man" (Oct. 1981). Examined, not reviewed by C. Aldous, 7/29/85. Useful interpretative summary of reproduction studies in rats and other species.

ADDITIONAL ANCILLARY REPRODUCTIVE STUDIES

Following are abstracts of various rat reproduction studies and interpretive reports from 421-025: Rec. #s 037269-037298. (C. Aldous, 4/8/87)

NOTE: The studies in this file are generally summaries without individual data.

025 037271 Sept. 1981, "Study of the possible influence of compound EL-222 on fertility in the adult female Sprague-Dawley rat". In this study 10 dams were dosed with 35 mg/kg/day fenarimol or with acacia vehicle by gavage for 60 days after determination that they were cycling normally. Blood was sampled at proestrus after about 30 days of treatment, and luteinizing hormone and prolactin levels were unchanged. Cycle length was 4.3 days in controls vs 5.0 days in treated females (not statistically significant). Dams were mated with untreated males and laparotomized between day 13 and 17 of gestation. All 10 females in both groups became pregnant. The numbers of implanted fetuses and grossly observable abnormalities

were comparable between groups. Dams were allowed to continue to term. Only about 50% of fetuses were delivered live in controls or treated groups (losses attributed to laparotomy). Prolactin levels induced by suckling were not affected by treatment.

025 037274 Oct. 1981, "Effects of EL-222 on the binding of androgens to cytoplasmic and nuclear androgen receptors". Approximately 60-day old Wistar-strain males were castrated and allowed to recover for 7 days, during which time they were dosed with 350 ppm fenarimol or control diet. Animals were killed and various tissues were assayed in vitro for binding of a synthetic androgen to cytoplasmic or nuclear androgen receptors in the hypothalamic preoptic area, pituitary, prostate, and other tissues. There were no differences in cytoplasmic or nuclear binding of labeled synthetic androgen to any tissues tested. Concentrations of labeled androgen did not vary with dose in any tissues analyzed. In another segment of the report, pregnant dams were fed 350 ppm fenarimol or control diet from (presumably) day 3 or 4 of gestation to term. At 3 or 4 days of lactation, 8 female pups (reason for testing only females not clear) were administered [3H]-testosterone and killed 2 hr later. There was no difference in binding of label to nuclear components of hypothalamic preoptic area + amygdala in the pups. Investigators concluded that test article was not an antiandrogen.

025 037279 Oct. 1981, "A special fertility study with EL-222 (Fenarimol) in the Wistar rat." 25 day old male rats were fed either 350 ppm fenarimol or control feed throughout the study. At 40 days of age and weekly thereafter, rats were placed for 16 hr with untreated females which had been hormonally primed to be in proestrus. Treatment ended at 82 days of age. There was a lag in the percentage of males which inseminated females, and in percentage of males which impregnated females. At the end of the dosing period, weights of testes, seminal vesicles, and prostate glands of treated and control animals were equivalent, however epididymides of treated animals were significantly lower in weight. Following a 36 day recovery period, there were no difference in weights of any of these tissues.

025 037282 Oct. 1981, "Effects of EL-222 on the concentration of estrogen receptors and the conversion of testosterone to estrogens in the hypothalamus". Pregnant Wistar strain dams were fed control diet or 350 ppm fenarimol from (apparently) day 3 or 4 of gestation through

gestation. On postpartum day 3 or 4, some pups were killed and nuclear estrogen receptor assays were performed in hypothalamic preoptic area + amygdala nuclear preparations. Male pups of treated dams had markedly lower receptor levels than controls. Groups were small (n = 4), hence soundness of the data is not fully established. This result is consistent with the investigator's conclusions that inhibition of aromatase enzyme levels in brain inhibited the process of estradiol formation, which is an essential mediator in male sexual behavioral development. In another segment of the study, dams were dosed (presumably) beginning from 18 days after mating onward. On postnatal day 3 or 4, 12 females per group were administered [3H]-testosterone (route unspecified) and sacrificed. Levels of labeled estrone and estradiol were 5 to 10 times higher in controls than in treated animals. Sample sizes were small (n = 4 pooled samples/group).

025 037288 Jan., 1982, "Preparturition progesterone levels in fenarimol (EL-222) treated rats". This study primarily shows that Sprague-Dawley dams fed 350 ppm fenarimol which had delayed parturition also had delays in the precipitous drop in levels of progesterone, which normally precedes parturition. Investigators cited references indicating that marked reductions of this hormone are important to actuate parturition in rats, but not in humans, other primates, and guinea pigs.

025 037291 Jan, 1982, "Effect of prepuberal and prolonged administration of EL-222 on sexual behavior, serum LH and prolactin concentrations, and testicular, pituitary, adrenal, seminal vesicle, epididymal and penile weights in male rats". Male Wistar rats were administered 0 or 35 mg/kg fenarimol subcutaneously in corn oil daily from age 23-123 days. Ovariectomized females were hormone-primed to be receptive, and males were placed with such females weekly. Records were kept of the numbers of males displaying sexual behavior (criteria not given for such "display"). Percentages displaying sexual behavior were 9-41% in treated animals vs a consistent 95% in controls.

025 037298 Aug. 1984, "A study to compare the effects of EL-222 (Fenarimol, compound 56722) and EL-228 (Nuvarimol, Compound 44828) on parturition and the organization of male sexual behavior in the Wistar rat". Females were mated and placed on control diet or 350 ppm

fenarimol in feed. Parturition for fenarimol rats averaged 40.5 hr later than controls (statistically significant). Dystocia was common in treated dams. Only 52% of fenarimol dams delivered (compared to 90% in controls). Only 4 of the 12 litters born to fenarimol group dams survived 4 days. On approx. day 4, pups were sacrificed and nuclear estrogen receptor assays were performed in (hypothalamus preoptic area + amygdala) samples (tissues of 2-3 litter mates/sex pooled per experimental unit). A marked and statistically significant reduction in estrogen receptor concentration was seen in male pups of the fenarimol group.

REPRODUCTION STUDIES WITH GUINEA PIG:

025 037297 "A two-generation reproduction study with fenarimol (EL-222, Compound 56722) in guinea pigs". Lilly Research Labs, Dec. 1983. Fenarimol, 98.5%, 0 or 400 ppm (approx. 33-35 mg/kg/day) in diet. No parental NOEL (minor male weight gain decrements at 400 ppm). Reproductive NOEL = 400 ppm. Complete, but not acceptable as an independent reproduction study: only one dosage level tested, and that dosage was not proved to be at or sufficiently near to an MTD to be fully justified. The study was well executed, and did not demonstrate reproductive toxicity at a dosage level which caused severe reproductive effects in the rat. **Not acceptable** to independently fill reproduction data requirement, but useful and scientifically valid data. No additional information needed for this report. (C. Aldous, 4/8/86. One-liner updated without new worksheet, C. Aldous, 8/26/88).

034 042862 "A two-generation reproduction study with fenarimol in guinea pigs: Response to EPA comments dated August 14, 1984". Lilly Research, August 1984. Fenarimol, tech., 0 and 400 ppm in diet. New information was provided to dispel EPA concerns that fenarimol might have affected gestation survival in the above guinea pig reproduction study 025:037297. This slightly reduced gestation survival was noted in the review of that study, however this small difference was not flagged by DPR as a potential adverse effect. The new information therefore has no appreciable bearing on the status of the study. (Aldous, 8/15/86).

REPRODUCTION STUDIES WITH MOUSE:

024 037257 Reproduction, 3-generation, mouse, Lilly Research Labs, July, 1978. Fenarimol, tech. 0, 35, 70, and 140 ppm in diet. Reproductive effects NOEL = 70 ppm (increased stillbirths at 140 ppm). Parental NOEL = 140 ppm (HDT). Not complete, not acceptable (more information requested, however upgrade to full acceptability not possible). (Aldous, 2/21/86) (See under "Reproduction, Rat: Summary of rat reproduction data", p. 4 of this Summary of Toxicology Data, for discussion of aromatase inhibition on rats and mice).

REPRODUCTION STUDIES WITH RABBIT:

024 037261 "A cross-over fertility study with EL-222 in the Dutch-belted rabbit" [Study B7229], Lilly Research Labs, June, 1980. Fenarimol, Lot B30-C69-220, purity 97.7%. 0 and 35 mg/kg/day by gavage in 5% acacia solution. NOEL = 35 mg/kg/day (No apparent treatment-related parental or reproductive toxicity). Not acceptable to fill reproduction study data requirement, but useful data (study limited to a single generation, too few animals, only one apparently non-toxic dose employed). (C. Aldous, 3/26/86).

TERATOLOGY, RAT

Study 026 037300 and a major follow-up study (027-028 037310 & 037313) should be evaluated together to derive maternal and developmental toxicity NOELS of 13 mg/kg/day.

** 026 037300 "A teratology study with EL-222 in the rat, (Lilly #R06279) Lilly Research Labs, Jan. 1980. Fenarimol, tech. (97.9%). 0, 5, 13, and 35 mg/kg/day by gavage. Developmental toxicity NOEL = 13 mg/kg/day (increased hydronephrosis): developmental toxicity NOAEL = 35 mg/kg/day (HDT), based on results of subsequent studies (see 27/28:037310 and 037313, which demonstrate that hydronephrosis involving parenchymal damage is not persistent postnatally). Maternal toxicity NOEL = 35 mg/kg/day, in the context of the present study. The high dose is a reproductively toxic level in adult males and females (reduced fertility, dystocia associated with prolonged gestation and increased stillbirths) as demonstrated in several reproduction studies, therefore high dose selection is justified. **Acceptable.** (C. Aldous, 5/21/87). Study previously reviewed as unacceptable (Aldous, 4/10/86).

003 991731 Incomplete version of 026:037300, above. (C. Aldous, 7/26/85)

036 050761 Brief review presented as part of a rebuttal to DPR review of teratology studies 026:037300 and 027/028:037310 and 037313. Review entitled "An evaluation of apparent hydronephrosis in experimental animals treated with EL-222", 6/27/86. (Considered in 5/21/87 DPR review of 026:037300, above).

5 024 037267-037268 "A discussion of hydronephrosis as it relates to reproduction and teratology studies with EL-222". Discussion was examined on 3/31/86, but not cited in reviews of teratology studies. Conclusion was similarly to that expressed in 026:037300 and its supporting documents. C. Aldous, 9/12/88.

027-028 037310-037312 & 037313-037315 "The effect of prenatal fenarimol (EL-222, Compound 56722) exposure on kidney development and maturation in the rat" (Supplementary reproduction/teratogenicity study, rat), Lilly Research Labs, Nov. 1983. Fenarimol, tech. 99.9%. 0 and 35 mg/kg/day (gavage). No NOELs found. Effects at 35 mg/kg/day as follows: Prolonged gestation times and dystocia in females. Hydronephrosis, and misc. skeletal variants in pups or fetuses. Decreased newborn viability, and transitory increased neonate hypothermia. Increased resorptions. Contributes to acceptability of primary study 026:037300. Aldous, 4/11/86).

TERATOLOGY, RABBIT

** 070 089208 "A Developmental Toxicity Study of Fenarimol (EL-222 Compound 056722) Administered Orally to New Zealand White Rabbits," (D.G. Hoffman and D.R. Russell, Lilly Research Laboratories, Laboratory Project I.D. B00290, 10/29/90). Fenarimol (purity = 96.59%) was administered by gavage to mated New Zealand White rabbits (20/group) at 0, 15, 50 or 150 mg/kg/day on gestation days 6 - 18. **Maternal NOEL = 50 mg/kg** (There was an increased incidence in early resorptions, percent non-live implants and abortions, as well as a decreased mean percent litter size at 150 mg/kg. In addition decreased body weight gain and food consumption were observed at 150 mg/kg.) **Developmental NOEL = 50 mg/kg** (Fetuses showed a significant increase in supernumerary ribs, percent litters with malformed fetuses and percent fetuses with variations, as well as a decrease in percent normal fetuses per litter at 150 mg/kg.) **No adverse effects indicated.** Acceptable. M. Silva, 9/7/93.

026 037299 "A teratology study on compound 56722 (EL-222) in the rabbit", (Lilly #B-7125), Lilly Research Labs, Jan. 1977. Fenarimol, tech., 97.9%. 0, 3, 10, and 35 mg/kg/day. Apparent NOEL = 35 mg/kg/day (HDT) for maternal and developmental toxicity. Not complete, not acceptable. Dose levels not justified, and appear to be too low. (C. Aldous, 4/9/86, 5/21/87).

003 991730 Incomplete version of 026:037299 reviewed by C. Aldous, 7/26/85.

036 050763 "Pilot teratology study in rabbits with compound 56722 (EL-222, Fenarimol)" (Study B7075). This is pilot to study B-7125 = DPR #026:037299. Data consists of 3 tables: food consumption, body weight, and uterine and fetal exam data. Median food consumption slightly reduced in 35 and 100 mg/kg/day groups; no significant differences in body weights between groups; increase in cases of >1 resorption/dam in 100 mg/kg/day group, but **lower** percentage of dams with resorptions than other groups. Pilot study does not justify dosage range selected for primary study, rather it suggests that the dosage range should have been higher.

GENE MUTATION

Bacteria

** 057 070840 "The Effect of Fenarimol (EL-222, Compound 56722) on the Induction of Reverse Mutations in Salmonella typhimurium and Escherichia coli Using the Ames Test," (Lilly Research Laboratories, Project Id: 880215AMT4 and 880222AMS4, 8-24-88) Fenarimol technical (lot B30-C69-220; 97.9% pure) was tested in the Ames plate incorporation method with Salmonella typhimurium strains TA1535, TA1537, TA98 and TA100 and Escherichia coli WP2uvrA-. Cytotoxicity and Fenarimol precipitation tests were performed. Bacteria were exposed to 125, 250, 500, 1000 and 2000 ug/plate (no S-9 activation) and 62.5, 125, 250, 500 and 1000 ug/plate (with S-9). S-9 was obtained from livers of male Fischer 344 rats. All concentrations were plated in

triplicate. **No mutagenic effects were observed at any dose. Acceptable.** D. Shimer & M. Silva, 3/14/90.

003 991737 "The effect of Lilly compound 56722, EL 222, upon bacterial systems known to detect mutagenic events." (7/9/1979, Lilly). Salmonella strains TA1535, TA100, TA1537, TA1538 TA98 and others were exposed to EL 222 (lot no B30-C69-220, no purity stated) over 0.1-1000 ug/ml by the gradient plate method devised by Lilly. The technique is not adequately described and justified, no evidence that the limit of solubility or cytotoxicity was approached, diffusion of the test article in agar not addressed, no individual plate counts and the method does not lend itself to statistical analysis. A concern is that the S9 activation is added to a layer of agar over the one containing the test article. In order for the compound to reach the bacteria, it must diffuse through the agar containing the S9 while incubated at 37 degrees (where the enzymes have a limited half-life) and then to the bacterial streaks. **Unacceptable** (no repeat trial, no purity or characterization of test article, protocol.) No evidence of increased reversion rate. C. Aldous, 7/30/85

023 037250-037251 Exact duplicate of 003 991737 (retain all record numbers).

023 037246-037247 "The effect of a-(2-chlorophenyl)-a-(4-chlorophenyl-5-pyrimidinemethanol, EL 222, Lilly compound 56722, on bacterial systems known to detect mutagenic events." (1976, Lilly). Salmonella, eight strains and E. coli, two strains, exposed to EL222 (lot B30-69-21, no purity stated), at 0.01 to 100 ug/ml, with and without rat liver activation; used the gradient modification of the Ames assay which does not lend itself to statistical analysis (see above also); compound was incorporated into the lower "wedge" presenting a question of diffusion and actual exposure of the bacterial streaks. **Unacceptable** (no repeat trial, question of diffusion, no purity or characterization of the test article.) J. R. Gee, 4/16/86.

Mammalian cells

057 070839 "The Effect of Fenarimol on the Induction of Forward Mutation at the Thymidine Kinase Locus of L5178Y Mouse Lymphoma Cells" (Lilly Research Laboratories, Project Id: 880106MLT4 and 880113MLA4, 4-19-88). Fenarimol (lot B30-C69-220; 97.9% pure) was tested on L5178Y mouse lymphoma cells, with and without S-9 activation (from Aroclor induced Fischer 344 male rat livers) at 1, 10, 20, 30, 35, 40, 45 and 50 ug/ml (4 hour incubation). Mutagenicity was assessed from triplicate cultures. Preliminary cytotoxicity and Fenarimol precipitation tests were performed. **No adverse effect** (A mutagenic effect was not observed with Fenarimol in this study). **Not Acceptable** (no repeat trial). D. Shimer & M. Silva, 3/12/90.

003 991738 "Evaluation of Lilly compound 56722, Fenarimol, in the L5178Y TK+/- mouse lymphoma cell forward mutation assay." (8/1/1979, Lilly). Mouse lymphoma. Fenarimol (lot B30-C69-220, no purity stated); tested without activation at 0, 3, 6, 12, 25, 50 or 100 ug/ml, at 0, 3, 6, 12, 25 or 50 ug/ml with activation, 4 hours incubation; maximum concentration used is not justified on the basis of cytotoxicity with activation as the relative growth was not decreased adequately. A repeat trial is necessary in this study. Viability and mutation were measured after a 3-day expression time. Activation was provided by S9 from Fischer 344 rats. **Unacceptable** (single trial, inadequate evidence of cytotoxicity with activation.) No evidence of mutagenic activity reported. C. Aldous, 7/30/85.

EPA 1-liner: CORE grade - minimum. Non-mutagenic at 100 ug/ml with and without enzyme activation. [Note: Assuming this goes with Record # 991738, there is an error in that the highest concentration with S9 was 50 ug/ml.]

023 037252 Exact duplicate of 003:991738 (retain both record #s).

CHROMOSOMAL ABERRATION

** 063 073242 "Mutagenicity Test on 056722 In an in vitro Cytogenetic Assay Measuring Chromosomal Aberration Frequencies in Cultured Purified Human Lymphocytes," (Hazleton Laboratories, America, Study No. 10348-0-449, 8-26-88). Fenarimol technical (lot # B30-C69-220; 97.9% pure) was used on duplicate cultures of human lymphocytes at 0 (vehicle = DMSO), 1.75, 2.50, 6.25, 12.5, 17.5 and 25.0 ug/ml (no S-9) or 59.9, 79.8, 99.8, 130, and 160 ug/ml (+ S-9). Toxicity was manifested at the maximum concentration in both assays by a reduction in the number of mitotic cells as well as poor chromosome morphology. 100 cells/culture for the 4 highest concentrations were read. There was no significant increase in chromosomally aberrant cells at any concentration of Fenarimol. **No adverse effects. Acceptable.** D. Shimer & M. Silva, 3/14/90.

** 063 073243 "Mutagenicity Test on 056722 (EL-222) in the in vivo Rat Micronucleus Assay," (Hazleton Laboratories America, Inc., Study No. 10348-0-455, 10-28-88). Fenarimol technical (Lot #: B30-C69-220; 97.9% pure) was given in a single dose by gavage to Sprague-Dawley rats (5/sex/time point) at 0 (vehicle = corn oil), 125, 625 or 1250 mg/kg. Rats were then sacrificed at 24, 48 and 72 hours to examine erythrocytes for micronuclei (2 slides/animal were prepared; 1000 PCE/animal were scored). An MTD was achieved (1/15 males and 2/15 females died within 72 hours at 1250 mg/kg; clinical signs, primarily lethargy and mucus around the eyes, were observed at all dose levels in both sexes). No increase in micronuclei in bone marrow polychromatic erythrocytes was observed. **No adverse effects. Acceptable.** D. Shimer & M. Silva, 3/15/90.

**** 063 073244** "Mutagenicity Test on 056722 (EL-222) in the Rat Bone Marrow Cytogenetic Assay" (Hazleton, Study No. 10348-0-451, 1-27-89) Fenarimol technical (LOT #: B30-069-220; 97.9% pure) was administered in a single dose by gavage, to Sprague-Dawley (CRL:CD-BR) rats (5/sex/time point) at 0 (vehicle = corn oil) 125, 625 or 1250 mg/kg. Animals were sacrificed at 6, 18 or 30 hours to examine bone marrow cells for chromosomal aberrations (50 metaphase spreads/animal were read and mitotic index was based on at least 500 cells). Fenarimol was negative for inducing chromosomal aberrations under conditions of the assay. **No adverse effects. Acceptable.** D. Shimer & M. Silva, 3/15/90.

003 991739 "A dominant lethal study of compound 56722 (EL-222) in the rat." (1/1977, Lilly). Fenarimol (B30-C69-220, no purity stated), given in as single oral dose of vehicle or 350 mg/kg, to 10 per group of Harlan Wistar-derived male rats; mated 1:1 with females for 8 weekly periods; no signs of toxicity reported in treated males. Insufficient numbers of pregnant females were used to detect an effect, the single dose is not justified, no concurrent control as required or historical data with a positive control, no signs of toxicity for an MTD are reported, resorptions were not differentiated into "early" and "late". **Unacceptable** with no adverse chromosomal effect reported. C. Aldous, 7/29/85.

[Note: In a rebuttal submitted by the sponsor, this study was described as ancillary to the reproduction studies to answer a specific question and was not intended as a full-scale mutagenicity study.] J. R. Gee, 4/28/87.

EPA 1-liner: CORE grade - minimum. Non-mutagenic at single oral dose of 350 mg/kg.

023 037248 Exact duplicate of 003:991739 (retain both record #s).

**** 034 042860** "Test for mutagenicity of technical-grade fenarimol using a micronucleus technique in the mouse." (5/14/82, presumed C.E.R.T.I., Versailles, France, study no. 650). Mouse micronucleus test. Fenarimol, >98%; 10 males per group given 0 or 1 g/kg in two doses at 24-hour interval by oral gavage. Positive controls treated twice at 24 hr intervals with benzene. Sacrifices at 24, 48, and 72 hours for test article, 24 hr only for solvent and

positive controls. Statistically significant increase in polychromatic erythrocytes with micronuclei at 24 hr after the second dose (no effects at 48 or 72 hr). Initially reviewed as unacceptable but possibly upgradeable with submission of additional information with justification for using males only (guidelines require both sexes or satisfactory justification for variance), also need PCE/NCE ratio data for indication of the status of the bone marrow. In view of the **positive effect** and the discussion on the justification of one sex based on no difference in the toxicity between sexes (included in Document 421-036, page 47), the study is upgraded to **acceptable** with variations with the notation that the magnitude of the effect might be greater if a higher dose and/or females were included. J. R. Gee, 7/31/86 and 4/28/87.

EPA 1-liner: CORE grade - acceptable with positive for clastogenic effects in male mice at 1 gm/kg body weight at 24 hours. Unacceptable at 48 and 72 hours due to absence of concurrent controls.

046 060571 Three brief tables of historical data relevant to 034:042860. The only one of these given a record number is entitled "The induction of micronuclei as a measure of genotoxicity" [A report of the U.S. Environmental Protection Agency Gene-Tox Program], published in Mutation Research 123:61-118, 1983 [only historical data on pp. 69-71 were selected for submission]. The two pages preceding this publication are historical data from the laboratory of Dr. Siou, who conducted study 034:042860. These data are relevant because they represent incidence in male Swiss mice tested at the contract laboratory. In addition to the above historical data, two pages of historical data from Lilly Research Laboratories on ICR mice (mostly females) were included, immediately following the extracts from the EPA Gene-Tox publication. Only data from Dr. Siou's laboratory were considered satisfactory for historical reference for study 034:042860. There is no convincing evidence against the possible adverse effect noted in the initial review of the study. NOTE: The minutes of the meeting of DPR with Elanco on Nov. 24, 1987 indicate that a repeat study is planned and a final decision regarding possible adverse chromosomal effects will be made when that study is reviewed by Medical Toxicology. J. Gee and C. Aldous, 8/30/88.

034 042861 "Test for mutagenic potential of technical-grade fenarimol by examination for chromosomal damage in the Chinese hamster." (6/10/82, C.E.R.T.I., Versailles, France, study no. 658). Chromosomal aberrations, in vivo, Chinese hamsters. Fenarimol, >98%; 10 males per group given 2 doses of 250 mg/kg, vehicle control or MMS; treated groups sacrificed at 24, 48, or 72 hr; controls at 24 hr only; no evidence for aberrations; initially reviewed as **unacceptable** (use of males only was not justified, no evidence of marrow toxicity.) but upgradeable with justification of use of males only and report of mitotic indices or other evidence of marrow cytotoxicity. A rebuttal from the sponsor tries to justify the use of males by stating there is not evidence for sex differential in toxicity. There is no evidence, however, that the MTD was used or that the marrows were exposed. No change in status. J. R. Gee, 7/31/86 and 4/28/87.

EPA 1-liner: CORE grade - acceptable. Negative for mutagenic effects at doses of 250 mg/kg body weight times 2 in marrow cells of hamsters. [Note: this review is not consistent with the grade for Record #42860. Vehicle control sacrifices were done at 24 hours only so that the 48 and 72 hour test groups had no concurrent controls and to be consistent, should be unacceptable for those two time points.]

Summary: Study 034 042860 (1982) showed a positive effect in the micronucleus assay. Recently, however, the study was repeated (063 073243) using a dose range (high dose = 1250 mg/kg), rather than a single dose (1000 mg/kg--administered in 2 aliquots). In the repeat study, there were no positive effects, nor were there positive effects noted in any of the other studies, including the most recent (073242--chromosomal aberration in human lymphocytes and 073244--rat bone marrow cytogenetic assay). It is of particular note that there was no positive effects in human lymphocytes. Considering the results of all the assays, fenarimol does not produce an adverse effect in the form of chromosomal aberrations.

DNA DAMAGE

** 003 991740 "The effect of Lilly compound 56722 (EL-222) on the induction of DNA repair synthesis in primary cultures of adult rat hepatocytes." (6/1979, Lilly, Study 790503-1). Rat hepatocyte UDS. Primary rat hepatocytes were exposed to 0.05, 0.1, 0.5, 1, 5, 10, 50 and 100 nmoles/ml for five hours. Net nuclear grains for 20 nuclei for each concentration were reported. Cytotoxicity was noted at 50-100 nmoles. **Acceptable** with no increase in grain counts. C. Aldous, 7/29/85.

023 037249 Exact duplicate of 003:991740 (retain both record #s).

** 003 991729 "Evaluation of the carcinogenic potential of fenarimol (EL222) in the C3H/10T1/2 embryonic mouse fibroblast culture system." (8/1980, Lilly) Cell transformation. C3H/10T 1/2-clone 8 were exposed to 0, 4, 16, 128, 256 and 320 nmole/ml for 24 hours in growth medium containing 10% serum. After six weeks, foci were counted. Cultures with 320 nmole could not be evaluated due to extreme cytotoxicity. No type II and type III foci were found. **Acceptable** with no increase in transformation frequency. C. Aldous, 7/31/85.

023 037254 Exact duplicate of 003 991729 (retain both record numbers).

** 003 034780 "An evaluation of carcinogenic potentials of 56722 employing the C3H/10T1/2 cell transformation system." (6/28/1979, EG&G Mason). Cell transformation. Fenarimol (lot B30-C69-220, no purity stated); C3H10T1/2 cells were exposed for 18 hours to 8, 16, 32 and 64 ug/ml - higher concentrations were cytotoxic. Estimated molecular weight is 330 so the amount of test article in this study is in the same range as the one above (991729). One type III focus was found at 8 and 64 ug/ml, in 12 plates of each. Two type II foci were found at 32 ug/ml. The positive control, DMBA, gave much higher values. **Acceptable** with a positive response. In view of the lack of dose response and the low values, the positive finding is of questionable significance. C. Aldous, 7/30/85.

023 037253 Exact duplicate of 003:034780 (retain both record #s).

003 034779 "An evaluation of carcinogenic potential of compound 59156 employing the C3H/10T1/2 cell transformation system." (6/16/1979, EG&G Mason, study no 594-244-8) Cell transformation. Although this study is listed on the DPR spreadsheet for fenarimol, the report contains no positive identification of the test article. Therefore, until further identified, this study is not included in the final evaluation. No dose-related increase in transformation was reported. C. Aldous, 7/31/85.

Summary of cell transformation: Two studies on cell transformation were evaluated as acceptable. Because of the lack of dose response and the irreproducibility between labs of the transforming ability of fenarimol and the negative finding with DNA repair, the weight of evidence is that the one positive finding for cell transformation is not of biological significance.

NEUROTOXICITY

Not required at this time.